{rokbox title=|Overview of the developed PCR multiplex method for specific amplification and identification :: Image: Authors| thumb=|images/stories/ieo/imagenespublicaciones/centro-oceanografico-baleares-ieo-csic-molecular-marker-detection-natural-hybridization-pinna-nobilis-pinna-rudis-catanese-et-al-2024-thumb.jpg|}images/stories/ieo/imagenespublicaciones/centro-oceanografico-baleares-ieo-csic-molecular-marker-detection-natural-hybridization-pinna-nobilis-pinna-rudis-catanese-et-al-2024.jpg{/rokbox}

Gaetano Catanese, **Maite Vázquez-Luis**, Salvatore Giacobbe, José Rafael García-March, Maria Zotou, Prado Patricia, Orestis Papadakis, José Tena-Medialdea, Stelios Katsanevakis, Amalia Grau, 2024. **Internal transcribed spacer as effective molecular marker for the detection of natural hybridization between the bivalves *Pinna nobilis* and *Pinna rudis***

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Abstract: The *Pinna nobilis*, a Mediterranean mollusc, has suffered population declines due to a massive mortality event associated with various factors including the parasite *Haplosporidium pinnae*. Some populations show resilience, possibly due to local environmental conditions. In this study, a molecular multiplex PCR method was developed using species-specific primers targeting Internal Transcribed Spacer (ITS) regions of *P. nobilis* and *P. rudis*, allowing accurate species identification and hybrid detection. Samples from Mediterranean areas were analysed, including putative hybrids and individuals from five other bivalve species. DNA was isolated, ITS regions were amplified and sequenced, and phylogenetic analyses confirmed species differentiation and primer specificity. The multiplex-PCR successfully identified *P. nobilis*, *P. rudis*, and their hybrids based on distinct amplicon patterns. This study highlights the value of molecular tools in species conservation, especially for monitoring and managing hybridization, supporting effective biodiversity conservation strategies.

Keywords: DNA barcode, endangered species, hybrid detection, molecular tools, multiplex PCR, ribosomal unit